

ESTCP Cost and Performance Report

(ER-9914)



Demonstration of Bioaugmentation at Kelly AFB, Texas

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TABLE OF CONTENTS

	Page
1.0 EXECUTIVE SUMMARY	1
2.0 TECHNOLOGY DESCRIPTION	3
2.1 TECHNOLOGY DEVELOPMENT AND APPLICATION.....	3
2.2 PROCESS DESCRIPTION	3
2.3 PREVIOUS TESTING OF THE TECHNOLOGY	4
2.4 ADVANTAGES AND LIMITATIONS OF THE TECHNOLOGY.....	4
3.0 DEMONSTRATION DESIGN	7
3.1 PERFORMANCE OBJECTIVES	7
3.2 SELECTION OF TEST SITE(S).....	7
3.3 TEST SITE/FACILITY HISTORY/CHARACTERISTICS	7
3.4 PHYSICAL SET-UP AND OPERATION	9
3.5 SAMPLING/MONITORING PROCEDURES	9
4.0 PERFORMANCE ASSESSMENT	13
4.1 PERFORMANCE DATA.....	13
4.2 PERFORMANCE CRITERIA AND PERFORMANCE ASSESSMENT	14
4.3 TECHNOLOGY COMPARISON	19
5.0 COST ASSESSMENT.....	21
5.1 COST REPORTING.....	21
5.1.1 Microcosm Testing	21
5.1.2 Field Testing	21
5.2 COST ANALYSIS.....	23
5.2.1 Cost Comparison.....	23
5.2.2 Cost Drivers and Potential Cost Impacts	27
5.2.3 Cost Drivers	27
5.2.4 Life Cycle Costs.....	27
6.0 IMPLEMENTATION ISSUES	29
6.1 COST OBSERVATIONS.....	29
6.2 PERFORMANCE OBSERVATIONS.....	29
6.3 SCALE-UP AND OTHER SIGNIFICANT OBSERVATIONS.....	29
6.4 LESSONS LEARNED.....	30
6.5 END-USER ISSUES	30
6.6 APPROACH TO REGULATORY COMPLIANCE AND ACCEPTANCE.....	30
7.0 REFERENCES	31
APPENDIX A POINTS OF CONTACT	A-1

FIGURES

		Page
Figure 1.	Cross-Sectional Diagram of the Surface Geology at Site 360, Kelly AFB.....	8
Figure 2.	Cross Sectional Diagram of Test and Control Plots at Kelly AFB, TX.....	10
Figure 3.	Plan View of the Test Plot at Site 360, Kelly AFB.....	11

TABLES

		Page
Table 1.	Analytical Methods	12
Table 2.	Distribution of Chloroethene over Time	14
Table 3.	Performance Criteria for the Bioaugmentation Demonstration	15
Table 4.	Expected Performance and Performance Confirmation Methods.....	17
Table 5.	Estimated Cost of Microcosm Testing.....	22
Table 6.	Costs for Field Demonstration at Kelly AFB, TX	22
Table 7.	Costs for Field-Scale Demonstration	24
Table 8.	Costs Comparison for Field Demonstration at a Generic Site	25
Table 9.	Cost Comparison of Bioaugmentation and Permeable Reactive Barrier	26
Table 10.	Present Value Estimates for the Bioaugmentation Technology in the Biobarrier Configuration and Reactive Barrier.....	28

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ACRONYMS AND ABBREVIATIONS

AFB	Air Force Base
bgs	below ground surface
<i>c</i> -DCE	<i>cis</i> -1,2-dichloroethene
DO	dissolved oxygen
DoD	Department of Defense
ESTCP	Environmental Security Technology Certification Program
GC/FID-ECD	Gas Chromatograph/Flame ionization Detector-Electron Capture Detector
gpm	gallons per minute
MCL	maximum contaminant level
MTBE	methyl- <i>tert</i> -butyl ether
O&M	Operation and Maintenance
ORP	oxidation-reduction potential
PCE	tetrachloroethene
ppb	parts per billion
PPE	personal protective equipment
PVC	polyvinyl chloride
RNA	remediation by natural attenuation
RTDF	Remediation Technology Demonstration Facility
TCE	trichloroethene
U.S. EPA	United States Environmental Protection Agency
VC	vinyl chloride
VFA	volatile fatty acid
VOC	volatile organic compound

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Technical material contained in this report has been approved for public release.

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1.0 EXECUTIVE SUMMARY

Chlorinated ethenes such as tetrachloroethene (PCE) and trichloroethene (TCE) are some of the most common groundwater contaminants found at Department of Defense (DoD) facilities. In addition to their common presence, these compounds are persistent under most natural geochemical conditions at these contaminated sites. Remediation of these sites through biodegradation of the chlorinated ethenes is a promising alternative at many of the sites. Reductive dechlorination is the primary pathway for biodegradation of chlorinated solvents. With this pathway, the chlorine atoms on the ethenes are sequentially replaced by hydrogen atoms through a biologically-mediated process. Generally, the hydrogen is generated through fermentation of an electron donor. Although many microorganisms are capable of mediating the reductive dechlorination process, only *Dehalococcoides ethenogenes* is known to completely reduce PCE and TCE to ethene. Unfortunately, *D. ethenogenes* is not present at all chloroethene-contaminated sites and the reductive dechlorination process stalls at cis-1,2-dichloroethene (c-DCE). Under conditions such as these, the application of enriched cultures containing *D. ethenogenes* or closely related microorganisms is used to complete the reductive dechlorination process.

The primary objective of the demonstration was to determine if complete reductive dechlorination could be stimulated through the introduction of a culture known to contain halorespiring bacteria. Secondary objectives involved testing the robustness of the applied culture by depriving it of electron donor and adding sulfate to the system. Samples were collected at a frequency and analyses were performed to evaluate the objectives of the demonstration. The results of the chemical analyses indicated that the complete dechlorination was achieved through the addition of the microbial culture. Each of the performance objectives were met during the demonstration at Kelly Air Force Base (AFB). The data indicate that the KB-1 culture was capable of stimulating complete reductive dechlorination. In addition it was determined that the KB-1 culture was fairly robust with the elimination of the electron donor and the addition of the sulfate from/to the system.

In 1976, the United States Environmental Protection Agency (U.S. EPA) designated PCE and TCE as priority pollutants. The Safe Drinking Water Act Amendments of 1986 strictly regulate both of these compounds; each has a maximum contaminant level (MCL) in drinking water of 5 parts per billion (ppb) (U.S. EPA, 1996). When concentrations of these compounds at a contaminated site are too high, remedial action is required to lower the concentration and reduce the risk to human health and the environment.

Bioaugmentation was successfully demonstrated for achieving complete dechlorination at Kelly AFB where delivery of donor/nutrient amendments resulted in limited success. At Kelly AFB, dechlorination of PCE was demonstrated to hold up at c-DCE with only the addition of an electron donor. After the aquifer was augmented with KB-1, a prepared culture of halorespiring bacteria, complete dechlorination of PCE to ethene was observed.

Following the successful demonstration of the bioaugmentation technology, the robustness of the KB-1 culture was tested through the deprivation of electron donor and then the addition of sulfate. The objectives were to investigate the survivability of the KB-1 culture, evaluate any

residual dechlorinating activity, attempt to reestablish the level of activity to pre-shutdown levels, and to stress the culture by adding sulfate. After approximately one year without the addition of the electron donor, gene probe analysis on groundwater samples collected across the augmented test plot all tested positive for the presence KB-1, and none of the samples from the non-augmented control plot tested positive. Complete PCE dechlorination was observed in one well inside the test plot suggesting that the KB-1 culture was utilizing a source of electron donor already in the groundwater. After the addition of the electron donor, complete reductive dechlorination was quickly observed in all of the wells.

Sulfate was added to establish an initial in-situ concentration of 600 mg/L. A significant amount of the sulfate was reduced, decreasing the concentration to 50 to 60 mg/L within 6 weeks. No apparent impact on the dechlorination activity was observed from the added sulfate.

The implications from these data are that (1) the KB-1 culture was very robust being able to compete with, and survive among, the indigenous microbial population, and (2) bioaugmentation may not require continuous attention following inoculation at sites where the natural attenuation requirements are met.

2.0 TECHNOLOGY DESCRIPTION

2.1 TECHNOLOGY DEVELOPMENT AND APPLICATION

Chlorinated solvents are widely used as solvents, cleaners, and degreasing agents. As a result of spills and past disposal practices, these compounds are contaminants in groundwater, soil, and sediments. Standard remedial approaches have proven to be ineffectual and costly at removing these substances from the environment. Within the last 15 years, basic research on natural microbial dechlorination mechanisms has suggested that the destruction of chlorinated compounds may be practically achieved at some sites by stimulating bacterial reductive dechlorination in the field.

Stimulation of microbial reductive dechlorination is achieved through the injection of electron-donating substrates and nutrients into the groundwater to produce proper reducing conditions. While stimulated biodegradation of chloroethenes may be an effective method of site remediation at many sites, there are instances where complete degradation of PCE and TCE to ethene is not possible through the addition of electron donors alone. In these cases, the degradation of PCE and TCE stops at c-DCE or vinyl chloride, resulting in the accumulation of these degradation components. The partial dechlorination of PCE and TCE may be caused by the absence of dechlorinating microorganisms (i.e., dehalorespiring microorganisms).

Cultures that contain phylogenetically-related organisms to *D. ethenogenes* have been produced for the application in the field. Examples of such cultures include the Pinellas culture and the KB-1 culture. A field demonstration of the Pinellas Culture was conducted at Dover AFB, and indicated that the dechlorination of c-DCE to ethene occurred only after the addition of the culture.

2.2 PROCESS DESCRIPTION

For the technology demonstration, the bioaugmentation system was constructed as a plot that was hydraulically isolated. Hydraulic isolation of the plot was accomplished by recirculating water between one injection well and three extraction wells. To complete the installation of the test plots, one extraction well, three injection wells, and five monitoring wells were installed in an area of 20 ft by 30 ft.

The extracted groundwater was pushed into an equipment shed by the submersible pumps in the extraction wells, where the electron donors (methanol and acetate) were added to the groundwater stream to achieve a total concentration of 7.2 mM. The groundwater was then pumped back into the injection well. Groundwater recirculation rates were maintained near 3 gallons per minute (gpm) throughout the tests giving a residence time in the test cell of approximately 8 days.

In general, groundwater samples were collected every month during operation or when system operating parameters were modified. During each sampling event, groundwater was collected for volatile organic compound (VOC), volatile fatty acid (VFA), sulfate, nitrite, nitrate, bromide (tracer), and dissolved gas analyses. In addition, samples were collected for gene probe analysis for detection of the KB-1 culture. During the sampling, the groundwater was monitored for

several parameters in the field (i.e., pH, temperature, conductivity, dissolved oxygen (DO), oxidation-reduction potential, salinity, and turbidity). Groundwater sampling typically required 3 full days of labor for two technicians, but general operation and maintenance required daily monitoring of the system and collection of routine data.

The primary objective of the demonstration was to determine if complete reductive dechlorination could be stimulated through the introduction of a culture known to contain halorespiring bacteria. Secondary objectives involved testing the robustness of the applied culture by depriving it of electron donor and adding sulfate to the system. Samples were collected and analyses were performed at a frequency to evaluate the objectives of the demonstration. The results of the chemical analyses indicated that the complete dechlorination was achieved through the addition of the microbial culture.

Once the system has been installed, the labor requirements were relatively low. Daily monitoring of system operating conditions was required to ensure safe and consistent operation. With the system at Kelly AFB, fouling of the injection wells required regular surging and redevelopment of the wells. In addition, fouled recirculation tubing required replacement about every 3 or 4 months. In general, groundwater sampling was performed about every month or two during operation. Operation and monitoring of the system and sampling of the groundwater all could be performed in Level C personal protective equipment (PPE).

2.3 PREVIOUS TESTING OF THE TECHNOLOGY

Demonstration of the bioaugmentation technology for the in situ treatment of chlorinated ethenes has been conducted at several sites from bench-scale to field-scale application. Results of these demonstrations and tests range from failure to complete success. Often with the successful demonstrations, the results are not conclusive that the complete reductive dechlorination is directly result of the addition of the culture. A White Paper prepared for Environmental Security Technology Certification Program (ESTCP) presents the state of the technology along with case studies of the demonstrations that have been performed.

2.4 ADVANTAGES AND LIMITATIONS OF THE TECHNOLOGY

The advantages of bioaugmentation over traditional technologies for chlorinated solvent remediation, such as biostimulation or pump-and-treat, are cost and duration of cleanup project. Bioaugmentation is more cost effective than pump-and-treat technologies due to the high capital and operational costs of pump-and-treat systems. The installation and operation of the treatment system are the factors driving the cost of the pump-and-treat systems. Also, the duration of the remediation project may be shortened when bioaugmentation is used in place of a standard biostimulation process. The application of a culture to the contaminated aquifer likely would increase the biodegradation rates relative to simple biostimulation. Further, simple biostimulation may not achieve the remedial goals of complete reductive dechlorination to ethene.

The main advantages of bioaugmentation for remediation of chlorinated solvents include the following:

1. Bioaugmentation results in contaminant destruction, not simply phase transfer;
2. The technology utilizes the aquifer volume as an in situ bioreactor;
3. In situ destruction of the contaminant may relieve regulatory requirements associated with pumping followed by aboveground treatment;
4. In situ treatment minimizes water disposal and preserves water balance.

The main limitations of the bioaugmentation technology include the following:

1. The culture must establish a niche in the aquifer and be able to compete with the indigenous microorganisms for essential nutrients;
2. The application is limited to sites of sufficient permeability to allow manipulation of groundwater flow;
3. The overall effectiveness depends on the ability to distribute the culture adequately in the subsurface.

Bioaugmentation is an innovative technology and the status of regulatory acceptance is unknown.

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3.0 DEMONSTRATION DESIGN

3.1 PERFORMANCE OBJECTIVES

The primary objective of the demonstration was to determine if complete reductive dechlorination could be stimulated through the introduction of a culture known to contain halorespiring bacteria. Secondary objectives involved testing the robustness of the applied culture by depriving it of electron donor and adding sulfate to the system. Samples were collected and analyses were performed to evaluate the objectives of the demonstration. The results of the chemical analyses indicated that the complete dechlorination was achieved through the addition of the microbial culture. Each performance objective was met during the demonstration at Kelly AFB. The data indicate that the KB-1 culture was capable of stimulating complete reductive dechlorination. In addition, it was determined that the KB-1 culture was fairly robust with the elimination of the electron donor and the addition of the sulfate from/to the system.

3.2 SELECTION OF TEST SITE(S)

NAS Fallon was initially selected because a number of studies had been performed at the Site 1 location for reductive dechlorination and biostimulation. In addition, a test system was previously installed that could be used to conduct the demonstration. All of the studies were unsuccessful at achieving dechlorination to ethene. In some of these studies, the reductive dechlorination process could not be initiated.

After complete dechlorination could not be achieved at NAS Fallon in the microcosm tests using the Pinellas culture, it was decided that testing should be conducted at Kelly AFB, where bioaugmentation had successfully been demonstrated. At Kelly AFB, the objective was to determine the robustness of the KB-1 culture that was used at the site. At Kelly AFB, depriving the culture of electron donor for over a year would test the robustness of the culture. If the culture successfully rebounded and dechlorination was started again, the dechlorination process would be perturbed with the addition of sulfate to the test plot.

3.3 TEST SITE/FACILITY HISTORY/CHARACTERISTICS

The location for the demonstration is situated in the courtyard of Building 360. The demonstration site was selected for the original bioaugmentation study based on the presence and concentrations of the contaminants, access to an existing test infrastructure, hydrogeology/geology of site, site logistics (site access, electrical power, water, etc.). The site was selected for this demonstration because the existing infrastructure and data gathered to date provided the basis for the bioaugmentation study, and allows for additional studies to further enhance the understanding of the underlying principles of the technology and how various operational/environmental considerations impact the technology's performance.

The geology in the vicinity of the test site consists of unconsolidated alluvial deposits that have been deposited on the top of the undulatory erosional surface of the Navarro Clay (see Figure 1). The alluvial deposits consist of gravel, sand, silt, and clay, ranging in thickness from 20 to 40 ft.

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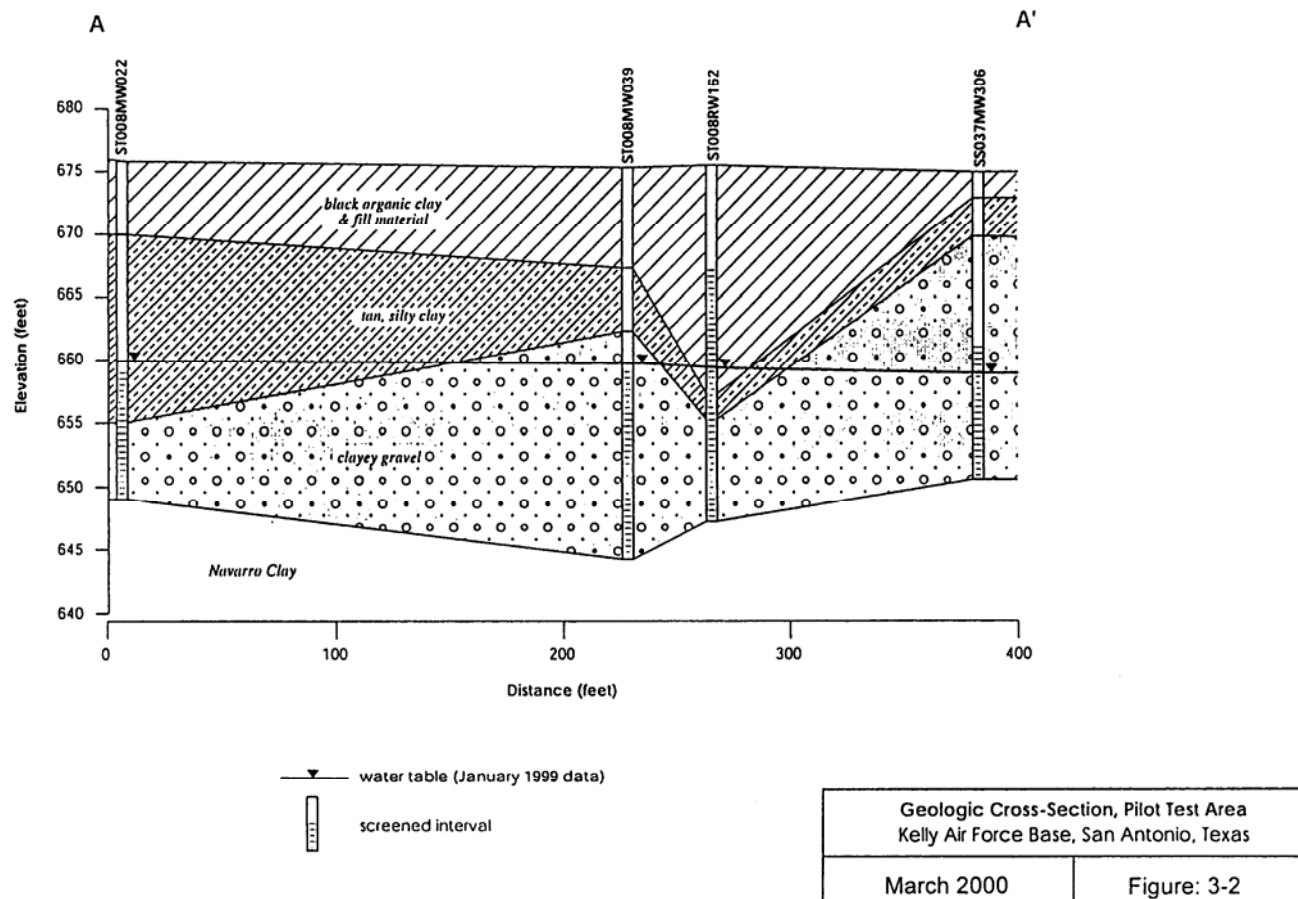


Figure 1. Cross-Sectional Diagram of the Surface Geology at Site 360, Kelly AFB.

From the surface downward, the geology typically consists of 1 to 4 ft of black organic clay, 6 to 16 ft of tan silty, calcareous clay; and 4 to 20 ft of clayey limestone and chert gravel (denoted as clayey/gravel). The surface of the Navarro Clay is irregular and characterized by ridges and channel-like depressions.

Groundwater in the area of the demonstration site is primarily present in the limestone/chert layer. The water table is approximately 15 to 20 ft below ground surface (bgs), and the saturated thickness is between 5 to 12 ft. Generally, groundwater flow is to the southwest with a flow velocity of approximately 0.3 ft/day. The regional water table gradient is approximately 0.003.

VOCs in the site groundwater consist primarily of PCE, TCE, and their degradation products c-DCE and vinyl chloride (VC). Total chlorinated ethene concentrations in the groundwater exceed 8,000 $\mu\text{g/L}$.

3.4 PHYSICAL SET-UP AND OPERATION

Each plot has a total of nine wells: one injection well, three extraction wells, and five monitoring wells. Figures 2 and 3 contain cross-sectional and plan views of the test systems, respectively. Three of the monitoring wells (B-X wells) are aligned along the center of the plot parallel to the groundwater flow direction and located at a distance of 8, 12, and 22 ft downgradient of the injection well. The other two monitoring wells (T-X wells) are aligned perpendicular to groundwater flow, and were initially installed to be outside the zone of influence of the system. Each of the wells in both plots are completed to a depth of 25 ft bgs and were screened from 15 to 25 ft to reduce the opportunity for aeration and increased oxygen concentrations of the groundwater as it moved through the treatment system.

An injection/extraction process was used hydraulically isolate the test and control plots. The injection/extraction rates were the same as those used during the Remediation Technology Demonstration Facility (RTDF)/GeoSyntec project (approximately three gpm). These injection/extraction rates were calculated by GeoSyntec using a groundwater modeling program and were demonstrated to have adequate isolation of the test cells and allow for a reasonable residence time in the cells during the RTDF/GeoSyntec project. Groundwater was extracted from the extraction wells using Grundfos submersible pumps and injected into the injection well after the addition of the amendments (electron donor, nutrients, etc.). The groundwater was pumped through a mobile shed where the nutrients were injected into the water stream using piston-style metering pumps.

Discussion of the operational conditions and periods of operation is presented in Section 4.1 (Performance Data) of this document.

3.5 SAMPLING/MONITORING PROCEDURES

Groundwater samples were collected throughout all phases of the demonstration to evaluate the performance of the bioaugmentation technology at the Kelly AFB site. A peristaltic pump was used to purge 3 well volumes of water out of each well. The purged groundwater was passed through an inline flow through cell and then into a waste container. While the water was being purged, a Water Quality Meter was placed inside the flow-through cell and was used to measure

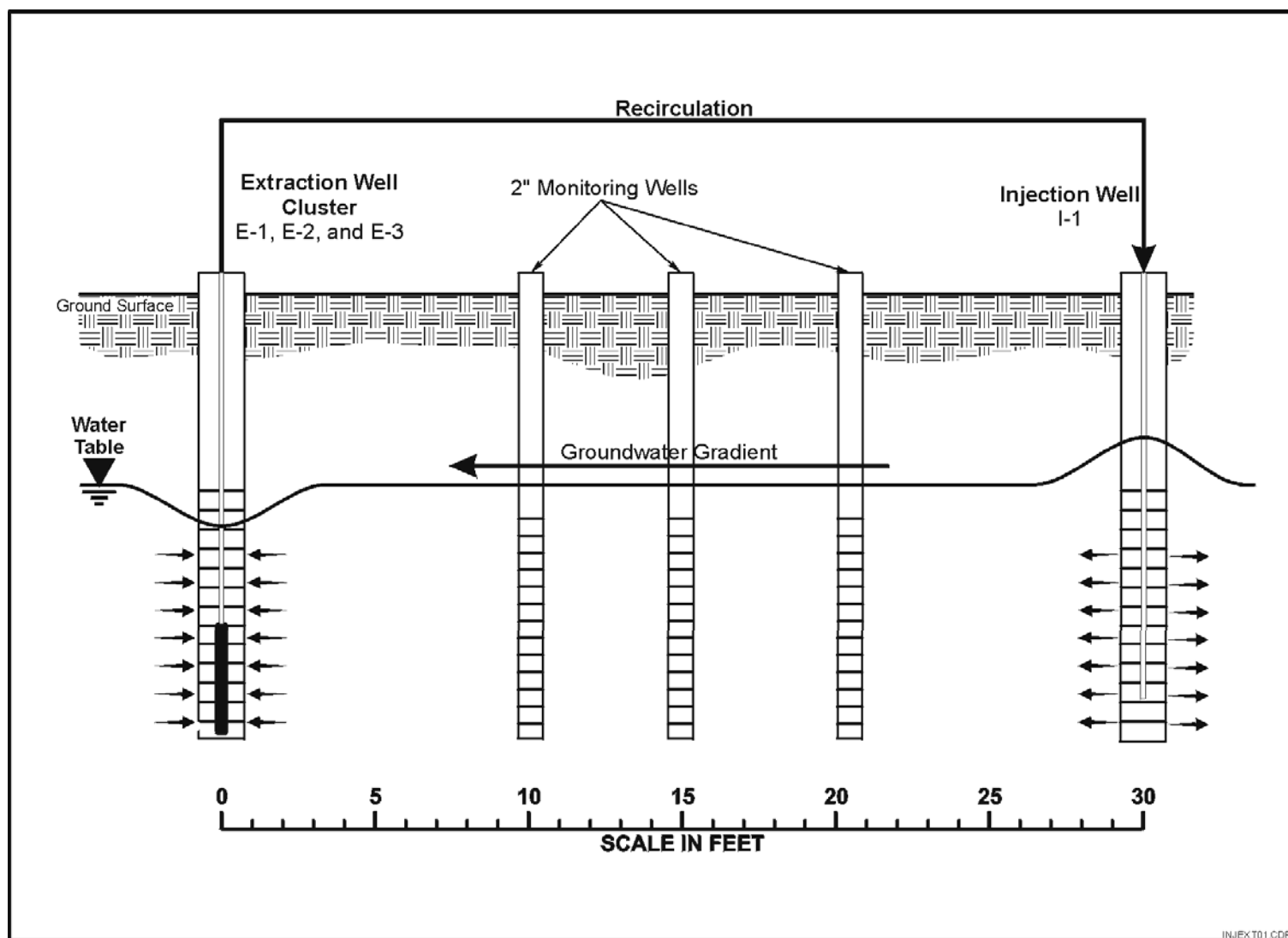


Figure 2. Cross Sectional Diagram of Test and Control Plots at Kelly AFB, TX.

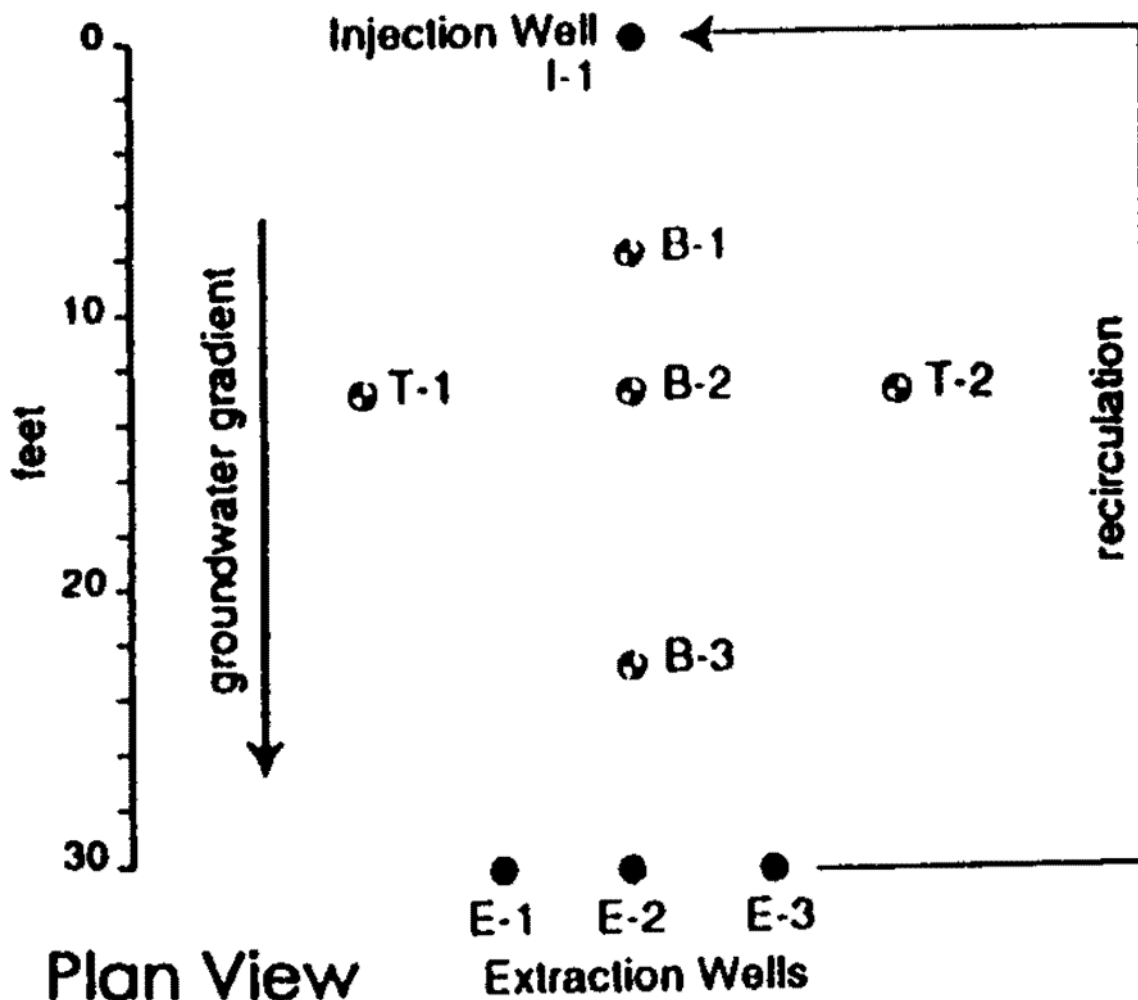


Figure 3. Plan View of the Test Plot at Site 360, Kelly AFB.

the pH, conductivity, turbidity, DO, temperature, salinity, and oxidation-reduction potential (ORP) of the water. Once the purged water was removed from the well, the appropriate bottles were used to collect the samples of water. The VOC samples were preserved with HCl, and the samples were then packed with ice in a cooler and shipped to Alpha Analytical Laboratories for analyses. A complete list of analyses, standard methods, hold times, and location of analysis is presented in Table 1.

The samples were analyzed both in the field and in the laboratory, depending on the specific parameter being measured (Table 1). Groundwater samples were collected prior to starting the system to obtain baseline analyses. These samples were analyzed at a laboratory for PCE, TCE, DCE, VC, ethene, ethane, methane, VFAs, bromide, nitrate, nitrite, and sulfate. Following the startup of the system, groundwater samples were collected to measure the effects of the

experimental parameters that were adjusted, and these samples also were analyzed for the laboratory and field parameters

In addition to the groundwater samples that were collected for chemical analyses, groundwater samples were collected to monitor the transport and survivability of the microbial culture through the test cells. These samples were collected prior to the start of the test to obtain baseline conditions. After the initiation of the demonstration, additional samples were be collected evaluate the migration and survivability of the microbial population during the test. The samples were sent to DuPont, for analysis using gene probe assaying to detect the culture.

Table 1. Analytical Methods

Measurement	Method	Instrumentation	Analysis Location
<i>Critical Measurements</i>			
PCE, TCE, <i>c</i> -DCE, VC,	SW 846 Method 8260B	Gas Chromatograph/ Flame Ionization Detector- Electron Capture Detector (GC/FID-ECD)	Laboratory
Ethene, Ethane, and Methane	EPA Standard Procedure (SOP)	GC FID	Laboratory
Volatile Fatty Acids (electron donor)	EPA (SOP)	GC/FID	Laboratory
Sulfate	EPA Method 300	Ion Chromatograph/ Conductivity Detector	
Bromide	EPA Method 300	Ion Chromatograph/ Conductivity Detector	Laboratory
<i>Non-critical Measurements</i>			
Nitrate, Nitrite, and Sulfate	EPA Method 300	Ion Chromatography/ Conductivity Detector	Laboratory
Bromide	Direct Reading	Bromide-Specific Electrode	Field
Dissolved Oxygen (DO)	Direct Reading	DO Probe	Field
pH	Direct Reading	pH Probe	Field
Conductivity	Direct Reading	Conductivity Meter	Field
Fe ⁺²	Hach Test Kit	Colorimeter	Field

4.0 PERFORMANCE ASSESSMENT

4.1 PERFORMANCE DATA

Table 2 presents the chloroethene and ethene molar distributions in percent (of the compound per total molar chloroethene/ethene concentration) over the duration of the testing at Kelly AFB. These data are the average concentration of each ethene species from every well that was sampled. Samples were collected from before the system was started until the system was turned off (after the sulfate was added to the test plot). Dates that the system conditions were modified are as follows:

Baseline sampling and start of the system	November 12, 1999
Start electron donor addition	February 9, 2000
Addition of culture	May 6, 2000
Stop electron donor addition	September 25, 2000
Die-off samples collected	August 23, 2001
Start addition of sulfate (3.6 mM)	March 9, 2002
Start addition of sulfate (7.2 mM)	July 19, 2002

The changes in the chloroethene distribution relative to the modification in system operating conditions demonstrate the effect of the modification on the reductive dechlorination potential. The baseline distribution of the chloroethenes (11/12/99) indicated that PCE was the dominant chloroethene species and that limited reductive dechlorination was occurring through the presence of c-DCE. Following the addition of the electron donor, the chloroethene concentrations are affected by limited reductive dechlorination (i.e., the PCE concentrations decrease while the c-DCE concentrations increase). Complete dechlorination does not occur until after the test plot was bioaugmented on May 6, 2000. Within 72 days of the addition of the culture, ethene is detected in the test plot and the PCE, TCE, and c-DCE are near the lowest levels observed during the demonstration. These data indicate that the addition of the KB-1 culture promoted complete reductive dechlorination.

After demonstrating the effects of bioaugmentation for the potential to promote complete reductive dechlorination, the system was shut down and the addition of the electron donor was stopped on September 25, 2000. Groundwater samples were collected from the test plot on August 23, 2001 to determine the effects of eliminating the electron donor for one year on the populations of the KB-1 culture and the reductive dechlorination process. The microbial analyses and the distribution of chloroethenes indicated that the KB-1 culture was present and complete dechlorination was still occurring.

Sulfate was added to the system at 3.6 mM on March 9, 2002 to determine if the competitive use of the electron donor between the chloroethenes and sulfate would limit the reductive dechlorination occurring in the test plot. Data generated after May 9, 2002 indicate that the addition of sulfate did not significantly affect reductive dechlorination.

Table 2. Distribution of Chloroethene over Time

Date	Distribution of Chloroethene/Ethene (%)				
	PCE	TCE	c-DCE	VC	Ethene
11/12/99	72.5	1.6	25.7	0	0
2/15/00	73.0	1.3	25.6	0	0
3/16/00	68.6	2.6	28.7	0	0
5/3/00	16.3	1.4	82.3	0	0
5/22/00	21.5	11.4	66.5	0.5	0
6/5/00	18.4	19.1	62.4	0.1	0
6/27/00	12.7	2.7	83.0	1.6	0
7/17/00	10.2	0.7	76.3	8.4	4.4
8/7/00	10.0	0.6	32.5	15.9	41.0
8/29/00	10.7	0.5	20.7	8.9	59.2
9/25/00	9.0	0.4	10.2	4.0	76.5
8/23/01	21.3	1.8	45.8	17.5	13.5
10/11/01	8.6	0.8	70.9	19.8	0
11/7/01	19.8	0.8	14.4	9.6	55.4
11/28/01	15.3	0.8	18.3	9.5	56.1
12/18/01	16.9	0.9	19.0	8.9	54.3
3/19/02	7.9	1.2	40.9	50.0	NT
4/25/02	3.9	0.9	32.9	16.8	45.5

4.2 PERFORMANCE CRITERIA AND PERFORMANCE ASSESSMENT

Table 3 presents the criteria that were used to assess the performance of the technology during the demonstration. The performance criteria are defined as primary or secondary depending on the importance to evaluating the performance of the technology.

The effectiveness of the bioaugmentation technology at achieving complete dechlorination was achieved by comparing the results produced in the test plot to those generated from the operation of a control plot within the same plume. The operating conditions and electron donor addition were same for both the control and test plots. In addition, prior to the addition of the culture, the system was allowed to operate until steady-state conditions had been achieved.

As was done with the testing of the overall bioaugmentation technology, the effects of eliminating the electron donor and the addition of sulfate were examined with the comparison of the results in the test plot with those in the control plot. Steady-state conditions also were achieved prior to modifying the conditions (i.e., electron donor and sulfate addition) in the test plot.

Table 3. Performance Criteria for the Bioaugmentation Demonstration

Performance Criteria	Description	Primary or Secondary
Contaminant Reduction	This technology is designed to reduce chloroethene contamination through sequential dechlorination to produce ethene as a final product.	Primary
Contaminant Mobility	Through the sequential dechlorination process, the mobility of the products is not substantially increased or decreased.	Secondary
Hazardous Materials	If successful conducted, no hazardous materials would remain or be introduced through the implementation of the bioaugmentation technology. However, the use of bioaugmentation may prevent the formation and accumulation of more hazardous compounds, such as vinyl chloride that may be produced during biostimulation.	Primary
Process Waste	The use of this technology does not produce any process waste.	Secondary
Factors Affecting Technology Performance	<p>The bioaugmentation technology is affected by groundwater geochemistry, hydrogeologic characteristics of the site, and survivability of the culture.</p> <p>Geochemistry: Sulfate inhibits the reductive dechlorination process. High or low pH, high salinity or high levels of metals may adversely affect the introduced culture.</p> <p>Hydrogeology: Low permeability may limit distribution of culture. High levels of organic matter may limit distribution of the culture, but may provide a source of electron donating substrate.</p> <p>Survivability of the Culture: Competition of the culture with the indigenous microbial population may affect the survival rate of the applied culture. Moderately alkaline conditions may favor the survival of the culture.</p> <p>Factors affecting the performance of the technology are discussed in greater depth in the Current State of the Bioaugmentation Technology.</p>	Primary
Reliability	The bioaugmentation technology as it was applied during the demonstration was relatively reliable. Problems were encountered with the recirculation pumps. However, this style of pumping would be eliminated during full-scale operation	Secondary
Ease of Use	Both at the demonstration-scale and with full-scale operation the technology is relatively easy to use. The only pieces of equipment that are used are pumps for the injection of electron donor.	Secondary

Table 3. Performance Criteria for the Bioaugmentation Demonstration (continued)

Performance Criteria	Description	Primary or Secondary
Versatility	This technology is likely very versatile depending on the culture applied and the target contaminant. Cultures have been produced to treat chloroethenes, MTBE, petroleum hydrocarbons, and chlorinated methanes, and PCBs. The bioaugmentation technology has long been used in the wastewater treatment systems.	Primary
Maintenance	Moderate maintenance was required for the technology demonstration. Daily monitoring of the system equipment and the water levels in the injection/extraction were required to ensure the injection well would not overflow and the water levels in the in the extraction well was not lowered beneath the top of the screen. Also, pumps, and electron donor solutions needed to be monitored to ensure continuous flow.	Secondary
Scale-Up Constraints	The widespread application of the culture represents the greatest challenge with the scale-up of the technology. Direct contact between the culture and the contaminant is imperative for success of the technology. As the culture is injected in a well, the contaminants are pushed in front of the microbial culture. Therefore, the use of an in situ biobarrier may be the most effective method to provide intimate contact between the contaminants and the culture.	Primary

A total of 15 sampling events were conducted over the course of the bioaugmentation study at Kelly AFB. In general, the sampling events occurred just prior to and then shortly after making a modification to the system test conditions. Following the sampling events near the modification, samples were collected about every month to investigate long-term effects of the system changes. During each sampling event, a complete suite of analyses was performed to determine the effects of the system modifications. For example, specific analyses were performed (i.e., microbial gene probe) to confirm the presence of the KB-1 culture in areas where complete dechlorination was occurring.

Table 4. Expected Performance and Performance Confirmation Methods

Performance Criteria	Expected Performance Metric (pre demo)	Performance Confirmation Method	Actual (post demo)
PRIMARY CRITERIA (Performance Objectives) (Qualitative)			
Contaminant Mobility	No change	Not measured	Uncertain
Faster Remediation	Achieve complete dechlorination and reduce remediation time	Monitor chloroethene concentrations in the test and control plots	The bioaugmented plot achieved complete dechlorination, while the control plot did not. Therefore, bioaugmentation would decrease remediation times relative to biostimulation and natural attenuation
Ease of Use	Minimal operator training required	Monitor labor requirements	Minimal operator training was required for continuous operation.
PRIMARY CRITERIA (Performance Objectives) (Quantitative)			
Feed Stream			
- Recirculation rate	2gpm	Continuous rotometer	Generally, achieved 2 gpm, but pump failure and water levels in the injection well reduced flowrates at times.
- Electron donor injection rate	3.6 mM (time-weighted)	Calibrated metering pumps	Achieved accurate injection levels
- Contaminant concentration	Total chloroethene 10 µM	U.S. EPA Method 8260	Maintained good mass balance
Target Contaminant			
- Percent reduction			
- Regulatory standard			
Hazardous Materials			
- Generated	None	Analysis for VC	Vinyl chloride was detected as a transient species
Process Waste			
- Generated	None	Observation	None detected
Factors Affecting Performance			
- Geochemistry	Geochemical conditions may limit survival of culture and dechlorination process	Analyze geochemical conditions (various methods), chloroethene concentrations (U.S. EPA Method 8260) and microbial populations	Natural water chemistry did not inhibit culture growth, nor did it prevent reductive dechlorination. Limited amounts of added sulfate (3.6 mM) did not affect dechlorination.

Table 4. Expected Performance and Performance Confirmation Methods (continued)

Performance Criteria	Expected Performance Metric (pre demo)	Performance Confirmation Method	Actual (post demo)
- Hydrogeology	Hydrogeologic conditions may limit distribution of culture	Performed tracer tests with microbial analyses	Hydrogeology at the site did not limit distribution of culture. The culture had populate the entire test cells within 3 months of injection
- Survivability	Lack of electron donor may kill culture	Eliminated electron donor addition, and monitored VFAs and microbial populations	This elimination of the electron donor addition did not stop reductive dechlorination process nor did the KB-1 culture die off
<i>SECONDARY PERFORMANCE CRITERIA (Qualitative)</i>			
Reliability	Limited shutdowns	Record Keeping	Moderate to high number of shutdowns due to pump failures, high groundwater levels, and fouling wells and tubing.
Safety			
- Hazards	Chloroethenes	Experience from demonstration operation	Level C PPE provided adequate protection
- Protective clothing	Level C personal protective equipment (PPE)		
Versatility			
- Intermittent operation	Yes	Experience from demonstration operation	Intermittent operation did not negatively affect system operation
- Other applications	Yes		Technology may be used for other chlorinated species and MTBE depending on the culture applied
Maintenance			
- Required	Regular changing of tubing, development of the injection well	Experience from demonstration operation	Regular replacement of the tubing was required and development of the injection well was performed, but fouling was still a problem.
- Eliminated	None		
Scale-Up Constraints			
- Distribution of Culture	Widespread distribution of culture would be required for large-scale application	Monitored migration of culture throughout demonstration	The culture was spread throughout the test plot relatively quickly due to the operation of the recirculation system. For large-scale application, the culture may need to be used in a biobarrier form to get intimate contact between the culture, electron donor, and contaminants

4.3 TECHNOLOGY COMPARISON

It is difficult to compare the performance of the bioaugmentation technology with other innovative alternative technologies, such as biostimulation. Bioaugmentation may be used to produce complete dechlorination that otherwise would not be achieved through simple biostimulation. From this standpoint, bioaugmentation may be the only option for meeting cleanup goals, if bioremediation is selected for the remedial action. Also, bioaugmentation may reduce cleanup times compared to biostimulation by eliminating the lag period between the initiation of electron donor injection and the onset of complete dechlorination, which may last several months in successful biostimulation projects. However, the time savings from using bioaugmentation is very difficult to predict. At some sites it may be less than one month at others it may be more than a year. The cost savings from reducing the lag time to achieve complete dechlorination is reduced by the cost of the culture and application of the culture. Because the reduction in the lag time is impossible to determine, it is difficult to compare bioaugmentation with biostimulation.

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5.0 COST ASSESSMENT

The bioaugmentation demonstration was conducted in two stages: a microcosm test (performed at NAS Fallon), and a full-scale demonstration conducted at Kelly AFB. The microcosm testing at NAS Fallon indicated the bioaugmentation would not be feasible at that site. Previous testing had been conducted at Kelly AFB; therefore, no microcosm testing was required prior to full-scale work at that site. Because microcosm testing is recommended prior to performing a full-scale remediation project, this cost assessment includes costing for both the microcosm and full-scale stages of the demonstration.

5.1 COST REPORTING

Throughout the course of this demonstration, the cost data were tracked to provide accurate cost information on the scale-up of the technology once it had been demonstrated. Costs associated with labor, consumable equipment, capital equipment (rented and purchased), subcontracted labor (Operation and Maintenance [O&M] providers), and purchased services (drillers and analytical) were tracked, and provide a basis for comparing bioaugmentation to other traditional technologies. Costs were tracked for both the microcosm and field-scale testing. The system used at Kelly AFB generally was established prior to the ESTCP testing at the site; therefore, some of the costs had to be estimated for the field scale testing of the technology.

The majority of the costs for bioaugmentation used to evaluate the cost performance of the system and to compare the costs of performing bioaugmentation to other technologies were obtained from the demonstration-scale tests. However, the majority of the system used during the demonstration had been constructed prior to the conduction of the demonstration, and these costs were estimated. For the full-scale implementation of the bioaugmentation technology, costs for the microbial culture were obtained from Regenesys, Inc. and the costs for microbial analysis were obtained from Sirem, Inc. Costs associated with performing pump-and-treat were estimated from previously performed projects. The majority of the costs for pump-and-treat were the installation of wells. Costs for performing the permeable reactive barrier technology were primarily obtained from the cost and performance report for this technology submitted to ESTCP.

5.1.1 Microcosm Testing

The cost to perform the microcosm testing option performed at NAS Fallon was estimated at \$78,000. Table 5 shows the cost breakdown. During the microcosm testing, two conditions were tested: an unaugmented control and augmented test bottles. Both of these conditions were conducted in triplicate and at least biweekly analyses were performed on the bottles. The soil samples were collected from an average depth of 20 ft bgs. Although GE provided the culture, an estimated cost of \$500 was used for GE to produce the culture.

5.1.2 Field Testing

The cost to complete a field test of bioaugmentation at Kelly AFB is presented in Table 6. The total cost of performing a field test of the bioaugmentation technology was estimated at \$255,936. Again, some of the costs associated with installation had to be estimated because the system had been used previously for bioremediation testing.

Table 5. Estimated Cost of Microcosm Testing

Activity	Unit Cost	Quantity	Cost
Microcosm Test Plan	\$5 K	1	\$5K
Microcosm Testing			
<i>Soil Collection</i>			
Labor	\$2K	1	\$2K
Travel	\$3K	1	\$3K
Drilling costs			
Mobilization	\$1K	1	\$1K
Drilling (20-ft deep)	\$25/lf	100 lf	\$2.5K
Waste disposal	\$2K	1	\$2K
Misc. (decontamination, etc.)	\$1K	1	\$1K
Consumables and supplies	\$1K	1	\$1K
	\$0.5K	1	\$0.5K
<i>Conduct Testing</i>			
Labor	15K	1	\$15K
Analytical services			
VOCs	\$100/sample	200	\$20K
Data analysis	\$5K	1	\$5K
Reporting	\$10K	1	\$10K
Total Cost for Microcosm Testing			\$78K

Table 6. Costs for Field Demonstration at Kelly AFB, TX

Cost Category	Subcategory	Costs (\$)
<i>FIXED COSTS</i>		
1. CAPITAL COSTS	Mobilization/demobilization	
	Mobilization of trailers	\$1,000
	Demonstration Plan	\$15,000
	Site work	\$20,000
	Equipment Cost	
	- Extraction/Metering Pumps	\$3,750
	- Manifold/Tubing	\$600
	Installation	
	- Drilling	\$22,367
	- Electrical	\$5,000
		Subtotal \$67,727
<i>VARIABLE COSTS</i>		
2. OPERATION AND MAINTENANCE	Labor	
	- Subcontractor	\$75,678
	- Battelle personnel	\$20,312
	Materials and Consumables	
	- Chemicals	\$3,000
	- Material	\$5,000
	Travel costs	\$9,250
	Culture	\$10,000
	Chemical/Biological Analyses	\$43,853
	Performance Data Analysis/Reporting	\$11,454
	Trailer Rental	\$9,600
		Subtotal \$188,209
TOTAL COSTS		
TOTAL TECHNOLOGY COST : \$255,936		

Note: Base provided electrical utility.

The layout of Kelly AFB consists of one injection well, three extraction wells, and six monitoring wells covering an area of approximately 30 ft by 20 ft. The total volume of groundwater treated by the demonstration system was approximately 40,000 gallons. Monitoring wells used for the demonstration were constructed of 2-inch polyvinyl chloride (PVC), and the injection and extraction wells were 4-inch PVC. The field trailers were used to store equipment and provided a location for the electron donor, tracer, and sulfate to be added to the system.

Mobilization costs included transporting the field trailers to the site and securing the trailers at the site. The majority of the site work costs include the construction costs for preparing the site, such as drilling and electrical installation. The labor and analytical costs are the dominant part of the variable costs, where the equipment and materials costs are much lower.

The estimated costs for performing the remediation effort at the scale of the demonstration is presented in Table 7. A cost of the remedial effort compared to the ESTCP demonstration indicates that the cost of the remedial effort would be approximately \$72,000 less than performing a standard demonstration. The fixed costs (system installation costs) would be nearly the same for both the demonstration and the remedial effort. However, the variable costs for the remedial effort would likely be lower than the standard demonstration because of the limited sampling and analysis. For the remedial project, samples could be collected on a quarterly basis. The labor costs decrease by about \$25,000 for the remedial effort because the routine maintenance would still be required for the remedial effort. However, the duration of the remedial project likely would be less for the remedial project due to fact that the remedial goals would be achieved faster than the performance goals of the demonstration.

5.2 COST ANALYSIS

5.2.1 Cost Comparison

A typical technology for treating chlorinated solvent-contaminated sites is pump-and- treat. Pump-and-treat is a traditional technology for remediating sites with chlorinated solvent contamination. For full-scale bioaugmentation operation, the use of a biobarrier would likely provide the most effective method of aquifer remediation. A comparison of the use of a biobarrier and pump-and-treat over time is provided in Table 8.

The costs presented in the cost comparison were derived from the generic site with a 5 acre chlorinated ethene plume having dimensions of 300 ft by 700 ft. The depth to groundwater is set as 15 ft and the total depth of the aquifer is 25 ft.

For construction of a biobarrier, it was believed that 20 wells would be required across the leading edge of the plume. Each of these wells would be screened across the thickness of the saturated zone. The biological culture would be injected into each of the wells, and the desired cell density (10^4 cells/ml) in the aquifer would be achieved through pumping and cell growth. It is estimated that approximately 25 L of the culture would need to be added to the system. The wells installed for the pump-and-treat system would be evenly spaced throughout the plume, and it was believed that 50 wells would be required to cover the plume.

Table 7. Costs for Field-Scale Demonstration

Cost Category	Subcategory	Costs (\$)
<i>FIXED COSTS</i>		
1. CAPITAL COSTS	Mobilization/demobilization	
	- Mobilization of trailers	\$1,000
	Work Plan	\$7,500
	Site work	\$20,000
	Equipment Cost	
	- Extraction/Metering Pumps	\$3,750
	- Manifold/Tubing	\$600
	Installation	
	- Drilling	\$22,367
	- Electrical	\$5,000
		Subtotal \$60,217
<i>VARIABLE COSTS</i>		
2. OPERATION AND MAINTENANCE	Labor	
	- Subcontractor	\$50,000
	- Battelle personnel	\$5,000
	Materials and Consumables	
	- Chemicals	\$3,000
	- Material	\$5,000
	Travel costs	\$5,000
	Culture	\$10,000
	Chemical/Biological Analyses	\$14,420
	Performance Data Analysis/Reporting	\$11,454
	Trailer Rental	\$9,600
		Subtotal \$113,474
TOTAL COSTS		
TOTAL TECHNOLOGY COST : \$173,691		

Note: Base provided electrical utility.

Table 8. Costs Comparison for Field Demonstration at a Generic Site

Cost Category	Subcategory	Bioaugmentation Costs (\$)	Pump-and-Treat Costs (\$)
<i>FIXED COSTS</i>			
1. CAPITAL COSTS	Mobilization/demobilization		
	- Mobilization of trailers	\$1,000	\$1,000
	Demonstration Plan	\$25,000	\$20,000
	Site work	\$20,000	\$100,000
	Equipment Cost		
	- Extraction/Metering Pumps	\$6,000	\$4,000
	- Manifold/Tubing	\$1,000	\$5,000
	- Treatment Equipment (Air Stripping/Catalytic Oxidizer)	\$0	\$105,000
	- Biological Culture	\$15,000	\$0
			\$15,000
	Installation		
	- Drilling with Disposal	\$33,000	\$83,000
	- Electrical	\$10,000	\$60,000
Subtotal		\$111,000	\$378,000
<i>VARIABLE COSTS</i>			
2. OPERATION AND MAINTENANCE	Labor (total)		
	- Subcontractor	\$130,000	\$390,000
	Materials and Consumables (total)		
	- Chemicals	\$48,000	\$0
	- Material	\$24,000	\$75,000
	- Electricity	\$5,000	\$25,000
	- Propane	\$0	\$20,000
	Chemical/Biological Analyses (total)	\$63,000	\$49,000
	Performance Data Analysis/Reporting	\$11,000	\$11,000
	Trailer Rental	\$10,000	\$10,000
	Subtotal	\$291,000	\$570,000
TOTAL TECHNOLOGY COST :		\$402,000	\$948,000
Quantity Treated: 814,000 gallons			

The costs for equipment and materials are much higher for pump-and-treat primarily because of the costs of the air stripping and catalytic oxidizer systems. It was estimated that these components would be approximately \$105,000. The only addition materials costs that bioaugmentation would have are with the biological culture (estimated at \$15,000).

The variable costs for pump-and-treat are significantly higher than those for bioaugmentation. While it is expected that the duration of the pump-and-treat system would be half as long as the biobarrier system, a significant cost associated with pump-and-treat is the operation and maintenance. It was estimated that the treatment systems for the pump-and-treat system would require 60 hours per/week while the bioaugmentation system would require 10 hours per week. The analytical costs associated with the biobarrier are only slightly higher due to the microbial analyses.

The total costs of the technologies would be \$402,000 for bioaugmentation and \$948,000 for pump-and treat. The total volume of groundwater that would be treated would be approximately

814,000 gallons. Therefore the unit treatment costs for bioaugmentation and pump-and-treat would be approximately \$ 0.50/gallon and \$1.16/gallon, respectively.

The costs of implementing bioaugmentation through the use of a biobarrier were also compared to the implementation of a permeable reactive barrier with iron medium. The cost associated with the permeable barrier were obtained from the cost and performance report for Evaluating the Longevity and Hydraulic Performance of Permeable Reactive Barriers at DoD sites (ESTCP, 2003). Costs for the permeable had to be estimated because unit costs were not presented in the report. It was assumed that the reactive barrier used during this cost estimate would need to be approximately 3 times as large as the barrier used during the field demonstration at NAS Moffet Field. The total cost of the sheet pile was estimated from the NAS Moffet Field installation. The NAS Moffet Field system was approximately 7.5 times narrower than the fictitious site used for these cost estimates. Because both technologies rely on natural groundwater movement, the treatment times for both the bioaugmentation and reactive barrier technologies were the same.

Table 9. Cost Comparison of Bioaugmentation and Permeable Reactive Barrier

Cost Category	Subcategory	Bioaugmentation Costs (\$)	Permeable Barrier Costs (\$)
FIXED COSTS			
1. CAPITAL COSTS	Mobilization/demobilization		
	- Mobilization of trailers	\$1,000	\$1,000
	Work Plan	\$25,000	\$25,000
	Site work	\$40,000	\$100,000
	Equipment Cost		
	- Extraction/Metering Pumps	\$6,000	\$0
	- Manifold/Tubing	\$1,000	\$0
	- Biological Culture	\$15,000	\$0
	Installation		
	- Drilling with Disposal	\$33,000	\$0
	- Electrical	\$10,000	\$0
	- Sheet Pile Installation	\$0	\$405,000
	- Reactive Barrier/iron medium	\$0	\$417,000
Subtotal		\$131,000	\$948,000
VARIABLE COSTS			
2. OPERATION AND MAINTENANCE	Labor		
	- Subcontractor	\$130,000	\$40,000
	Materials and Consumables		
	- Chemicals	\$40,000	\$0
	- Materials	\$24,000	\$0
	- Electricity	\$5,000	\$0
	Chemical/Biological Analyses	\$55,000	\$40,000
	Performance Data Analysis/Reporting	\$11,000	\$11,000
	Trailer Rental	\$10,000	\$10,000
Subtotal		\$275,000	\$101,000
TOTAL TECHNOLOGY COST :		\$406,000	\$1,049,000

Although the cost comparison in this report was made between bioaugmentation and pump-and-treat and bioaugmentation and permeable reactive barriers, a comparison may be made between bioaugmentation and biostimulation. However, a comparison between bioaugmentation and biostimulation is more difficult because the cost difference is not easily defined. The benefit

from applying a culture results from a potential decrease in remediation time, and the magnitude of this decrease is uncertain as well as site dependent. Therefore, the cost benefit from applying the bioaugmentation technology over biostimulation is uncertain.

5.2.2 Cost Drivers and Potential Cost Impacts

The costs provided for each testing option (i.e., microcosm or field test) were calculated under assumptions that were developed to describe a "typical" site. The actual costs for both microcosm testing and field testing would depend on site-specific requirements/ logistics. Due to the variability in site conditions, there is a large amount of uncertainty in the cost estimates used in this report. The variables that affect each approach and their potential impact are summarized in the following sections.

5.2.3 Cost Drivers

The single variable that could significantly impact the cost of conducting the microcosm tests is the depth of the contamination, which has a direct effect on the costs associated with collecting the aquifer core material, specifically the drilling, waste disposal, and labor costs. The costs presented in Section 5.1 assume a depth of 25 ft. Collection of cores from shallower sites would be somewhat less expensive, while collection of soil from deeper sites would obviously be greater. For example, if the contamination were located at 200 ft, the total cost of the microcosm test would increase on the order of \$40,000. The drilling costs would increase by \$22,000 and the disposal costs would increase by \$20,000.

The most significant cost variables for the field implementation of bioaugmentation are the hydraulic conditions at the site and the depth to the contamination. Lower hydraulic conductivities at a site would require a greater number of wells be used at the site to obtain relatively rapid distribution of the culture. Also, a greater number wells may be required to get even and rapid distribution of the electron donor and any nutrients. In general, a site with lower hydraulic conductivity would also require a longer period of operation, if the system relied on natural groundwater flow through the biobarrier, thus increasing operational costs. The impact that depth has on the costs, however, is much more pronounced than for the microcosm testing. Not only is the system installation cost impacted, but the cost of conducting the test is impacted as well. Implementing the bioaugmentation technology a 200-ft-deep site would result in a dramatic cost impact. The cost of labor for well installation would increase to \$90,000, and the waste disposal would increase to \$20,000. The materials costs would increase by four times due to install the system to the greater depth. The labor costs for conducting the test would increase primarily because of the need for increased sampling times.

5.2.4 Life Cycle Costs

For full-scale implementation of bioaugmentation, the capital costs and life-cycle costs are dependent on the design of the system used. As suggested previously, the most effective method of treating an aquifer with bioaugmentation likely would be a biobarrier. Capital costs for the installation of a biobarrier would be dependent on the depth of the aquifer and the lateral extent of contamination.

Operational costs would be relatively low due to the simplicity of the system. The bulk of operational costs would be associated with the regular sampling to ensure that the barrier is effectively treating the contaminated groundwater. Analysis would include chloroethene, dissolved gasses and VFA concentrations. The frequency of sampling and analysis would likely be dependent on the requirements of the overseeing regulatory agency.

Due to the relatively high capital cost for the installation of the biobarrier system, it would be recommended that microcosm or field treatability testing be performed prior to the full-scale implementation of the technology. If complete dechlorination to ethene is not observed in the microcosm or field-scale testing, full-scale operation of the technology should be reconsidered. Performing on small-scale testing should significantly reduce the liability associated with the partial dechlorination of PCE/TCE to another regulated compound, such as vinyl chloride.

Table 10 presents the life cycle costs for implementing the bioaugmentation technology in the biobarrier configuration and the reactive permeable barrier. For an operational period of 5 years, the total cost of the bioaugmentation technology would be \$816,000 and the reactive barrier would be approximately \$1,198,000. After 10 years of operation both technologies would be nearly the same at approximately \$1,500,000. If the systems operate 20 years and the barrier material has a life of 10 years, the total cost of the bioaugmentation technology would be \$2,871,000 and the reactive barrier would be 2,896,000.

Table 10. Present Value Estimates for the Bioaugmentation Technology in the Biobarrier Configuration and Reactive Barrier

Cost Scenario	Bioaugmentation	Reactive Barrier
Capital Investment Cost	\$131,000	\$948,000
Annual O&M Cost	137,000	50,000
Present Value over 5 years	816,000	1,198,000
Present Value over 10 years	1,501,000	1,448,000
Present Value over 20 years with 10 year life of barrier	2,871,000	2,896,000

6.0 IMPLEMENTATION ISSUES

6.1 COST OBSERVATIONS

Factors such as fouling of the injection well and transfer tubing in both the test and control plots affected the cost of the project at Kelly AFB. Fouling of these system components required additional maintenance costs such as redevelopment of the well and replacement of the tubing. In addition to the costs associated with the repair and replacement of the equipment, the downtime was costly to the project. The fouling of the system components was likely related to the geochemical conditions at the site. Likely oxidation of minerals in the groundwater during the extraction process and precipitation of the mineral in the injection well caused some of the fouling in the wells. Biological growth likely also resulted in some of the fouling of the wells. During full-scale operation, groundwater would not be recirculated, reducing the fouling potential of the wells in the biobarrier. However, for future projects that use a recirculation process, fouling is a potential.

6.2 PERFORMANCE OBSERVATIONS

The primary objectives of increasing remediation rates compared to biostimulation were achieved with the complete dechlorination that was accomplished with bioaugmentation and the incomplete dechlorination in the control (biostimulation) process. Also, no hazardous materials were produced (accumulated) with the bioaugmentation process; complete dechlorination to ethene occurred. With the complete dechlorination of PCE to ethene, regulatory objectives would have been achieved, and the migration of the contaminant would be minimized. The elimination of the electron donor and the addition of sulfate demonstrated that the added culture were relatively hardy and resistant to perturbation of the aquifer geochemistry.

The objectives of continuous operation were partially achieved. When the system was operating, the groundwater recirculation rates and rates of electron donor addition were relatively constant. However, fouling of the wells caused downtime in the operation.

6.3 SCALE-UP AND OTHER SIGNIFICANT OBSERVATIONS

Moving the bioaugmentation technology from demonstration-scale to full-scale implementation would require a different application of the technology. As mentioned previously, the full-scale implementation would involve the use of a biobarrier.

The greatest challenges to the successful implementation of a full-scale bioaugmentation project would be the adequate distribution of the microbial culture and the survival of the culture. Proper distribution of the microbial culture is dependent on the physical properties of the aquifer and the application of the culture. In general, more permeable aquifers and greater injection pressures enhance the distribution of the culture. The survival of the culture is primarily dependent on the compatibility of the culture with chemical and biological conditions of the aquifer. The survivability and distribution affect the feasibility of technology more than costs of implementation.

6.4 LESSONS LEARNED

With this demonstration, the bioaugmentation technology was demonstrated to be effective at reducing PCE to ethene at Kelly AFB. While other tests of the bioaugmentation technology have been performed at other locations and with other contaminants, the technology is very site and contaminant specific. The technology also proved unsuccessful at the proposed test site (NAS Fallon). Therefore, additional testing is required for the technology and certainly microcosm testing should be performed at a proposed site prior to conducting full-scale operation.

6.5 END-USER ISSUES

The design and application of bioaugmentation technology consists of installing or using simple components that are readily available. This technology however requires the introduction of organisms specifically selected/grown to operate in the subsurface environments where native organisms either are absent or are not robust enough to be simply biostimulated. Initial design and installation of a bioaugmentation system would require some specialized knowledge and it is the express purpose of the Remediation Technologies Development Forum (RTDF) to educate the public with respect to the knowledge needed to appropriately choose such a technology. They can be reached at: <http://www.rtdf.org/public/bioremed/biodocsp.htm>. Several documents have been placed there to assist remedial program managers.

6.6 APPROACH TO REGULATORY COMPLIANCE AND ACCEPTANCE

If the technology were implemented in the form of a biobarrier, the regulatory approval to conduct a full-scale bioaugmentation project would likely be limited to underground injection permits (for the culture and the electron donor). Generally, the underground injection permits are authorized by state regulatory agencies. Due to the minimal hazards associated with both the cultures and the electron donating substrates, regulatory approval is likely to be relatively quick.

7.0 REFERENCES

1. United States Environmental Protection Agency, 1996. *Drinking Water Regulations and Health Advisories*. EPA/822-3-96-002. Office of Water, Washington, D.C. October.

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APPENDIX A

POINTS OF CONTACT

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